

FIG. 1

DNA sequence for human
preproparathyroid hormone.

10 30 50
ATGATHCCNGCNAARGAYATGGCNAARGTNATGATHGNTATGYTNGCNATHGTGYTTYTN

70 90 110
ACNAARWSNGAYGGNAARWSNGTNAARAARMGNWSNGTNWSNGARATHCARYTNATGCAY

130 150 170
AAYYTNGGNAARCAYYTNAAYWSNATGGARMGNGTNGARTGGYTNGMNAARAARYTNAR

190 210 230
GAYGTNCAYAAYYTTYGTNGCNYTNGGNGCNCNYTNGCNCNCMNGAYGCNGGNWSNCAR

250 270 290
MGNCCNMGNAARAARGARGAYAAYGTNYTNGTNGARWSNCAYGARAARWSNYTNGGNGAR

310 330
GCNGAYAARGCNGAYGTNAAYGTNYTNACNAARGCNAARWSNCARTR

M = A or C
R = A or G
W = A or T
S = C or G
Y = C or T
H = A or C or T
N = A or G or C or T.

00520-1578960

FIG. 2

DNA sequence for human
preproparathyroid hormone in plasmid pSSHPTH-10.

10 30 50
ATGATGATACCTGCAAAAGACATGGCTAAAGTTATGATTGTCATGTTGGCAATTTGTTTT

70 90 110
CTTACAAAATCGGATGGGAAATCTGTTAAGAAGAGATCTGTGAGTAAAATACAGCTTATG

130 150 170
CATAACCTGGGAAAACATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCTG

190 210 230
CAGGATGTGCACAATTTTGTGTCCTTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTCC

250 270 290
CAGAGGCCCCGAAAAAAGGAAGACAATGTCTTGGTTGAGAGCCATGAAAAAAGTCTTGGA

310 330
GAGGCAGACAAAGCTGATGTGAATGTATTAATAAGCTAAATCCCAGTGA

FIG. 3

Portion of DNA sequence of the plasmid
for insertion into E. coli, coding for human
preproparathyroid hormone with flanking sequences.

```

      10              30              50
TATGATGATHCCNGCNAARGAYATGGCNAARGTNATGATHGTNATGYTNGCNATHGTGTT

      70              90              110
YYTNACNAARWSNGAYGGNAARWSNGTNAARAARMGNWSNGTNWSNGARATHCARYTNAT

      130             150             170
GCAYAAYYTNGGNAARCAYYTNAAYWSNATGGARMGNGTNGARTGGYTNMGNAARAARYT

      190             210             230
NCARGAYGTNCAYAAYYTTYGTNGCNYTNGGNGCNCNYTNGCNCNMGNAYGCNNGNWS

      250             270             290
NCARMGNCCNMGNAARAARGARGAYAAYGTYTNGTNGARWSNCAYGARAARWSNYTNGG

      310             330             350
NGARGCNGAYAARGCNGAYGTNAAYGTYTNACNAARGCNAARWSNCARTRRAAATGAAA

      370             390             410
ACAGATATTGTCAGAGTTCTGCTCTAGACAGTGTAGGGCAACAATACATGCTGCTAATTC

      430
AAAGCTCTATTA

```

M = A or C
R = A or G
W = A or T
S = C or T
Y = C or T
H = A or C or T
N = A or G or C or T.

005260-15109960

FIG. 4

DNA sequence for human preproparathyroid hormone in plasmid pSSHPTH-10 with flanking sequences.

10 30 50
TATGATGATACCTGCAAAAGACATGGCTAAAGTTATGATTGTCATGTTGGCAATTTGTTT

70 90 110
TCTTACAAAATCGGATGGGAAATCTGTTAAGAAGAGATCTGTGAGTGAAATACAGCTTAT

130 150 170
GCATAACCTGGGAAAACATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCT

190 210 230
GCAGGATGTGCACAATTTTGTGCCCCTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTC

250 270 290
CCAGAGGCCCCGAAAAAAGGAAGACAATGTCTTGGTTGAGAGCCATGAAAAAAGTCTTGG

310 330 350
AGAGGCAGACAAAGCTGATGTGAATGTATTAATAAGCTAAATCCCAGTGAAAAATGAAA

370 390 410
ACAGATATTGTCAGAGTTCTGCTCTAGACAGTGTAGGGCAACAATACATGCTGCTAATTC

430
AAAGCTCTATTA.

FIG. 5

DNA sequence coding for
preproparathyroid hormone in pSSHPTH-10 with flanking
sequences, showing the corresponding amino acid
sequence of preproparathyroid hormone.

10 30 50
TATGATGATACCTGCAAAAGACATGGCTAAAGTTATGATTGTCATGTTGGCAATTTGTTT
MetIleProAlaLysAspMetAlaLysValMetIleValMetLeuAlaIleCysPh

70 90 110
TCTTACAAAATCGGATGGGAAATCTGTTAAGAAGAGATCTGTGAGTGAAATACAGCTTAT
eLeuThrLysSerAspGlyLysSerValLysLysArgSerValSerGluIleGlnLeuMe

130 150 170
GCATAACCTGGGAAAACATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCT
tHisAsnLeuGlyLysHisLeuAsnSerMetGluArgValGluTrpLeuArgLysLysLe

190 210 230
GCAGGATGTGCACAATTTTGTGGCCCTTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTC
uGlnAspValHisAsnPheValAlaLeuGlyAlaProLeuAlaProArgAspAlaGlySe

250 270 290
CCAGAGGCCCGAAAAAAGGAAGACAATGTCTTGTTGAGAGCCATGAAAAAGTCTTGG
rGlnArgProArgLysLysGluAspAsnValLeuValGluSerHisGluLysSerLeuGl

310 330 350
AGAGGCAGACAAAGCTGATGTGAATGTATTAATAAGCTAAATCCCAGTGAAAATGAAA
yGluAlaAspLysAlaAspValAsnValLeuThrLysAlaLysSerGlnEnd

370 390 410
ACAGATATTGTCAGAGTTCTGCTCTAGACAGTGTAGGGCAACAATACATGCTGCTAATTC

430
AAAGCTCTATTA.

Figure 6. Nucleotide sequence of the MF 1-HPTH fusion gene from pS LX5-HPTH1. Nucleotide nos. 1-173 make up the MF 1 promoter region and 5' noncoding sequence. 174-440 is the MF 1 N-terminal coding sequence. 441-695 is the HPTH sequence obtained from pSSHPTH-10. 696-726 is an HPTH 3' noncoding sequence from pSSHPTH-10. 727-732 is from pUC19. 733-874 is MF 1 3' noncoding sequence and transcriptional termination signal.

```

10      10      30      50
AGTGCAAGAAAACCAAAAAGCAACAACAGGTTTTGGATAAGTACATATATAAGAGGGCCT

      70      90      110
TTTGTTCCTCATCAAAAATGTTACTGTTCTTACGATTCATTTACGATTCAAGAATAGTTCA

15      130      150      170
AACAAGAAGATTACAACTATCAATTCATACACAATATAAACGACCAAAAAGAATGAGAT

      190      210      230
TTCCTTCAATTTTTACTGCAGTTTTATTTCGCAGCATCCTCCGCATTAGCTGCTCCAGTCA

      250      270      290
ACACTACAACAGAAGATGAAACGGCACAAATTCCGGCTGAAGCTGTCATCGGTTACTCAG

20      310      330      350
ATTTAGAAGGGGATTTTCGATGTTGCTGTTTTGCCATTTTCCAACAGCACAAATAACGGGT

      370      390      410
TATTGTTTATAAATACTACTATTGCCAGCATTGCTGCTAAAGAAGAAGGGGTATCTTTGG

      430      450      470
ATAAAAGAGAGGCTGAAGCTTCTGTGAGTGAAATACAGCTTATGCATAACCTGGGAAAAC

25      490      510      530
ATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCTGCAGGATGTGCACAATT

      550      570      590
TTGTTGCCCTTGAGAGCTCCTCTAGCTCCCAGAGATGCTGGTTCCCAGAGGCCCCGAAAAA

      610      630      650
AGGAAGACAATGTCTTGTTGAGAGCCATGAAAAAAGTCTTGAGAGGCAGACAAAGCTG

      670      690      710
ATGTGAATGTATTAATAAGCTAAATCCAGTGAAAATGAAAACAGATATTGTCAGAGT
5

```

005250" 75785950

730 750 770
TC1 TTAGAGTCGACTTTGTTCCCACTG TTAGCTCGTACAAAATACAATATAC
90 810 830
TTTTCAATTTCTCCGTAAACAACCTGTTTTCCCATGTAATATCCTTTTCTATTTTTCGTTT
850 870
CGTTACCAACTTTACACATACTTTATATAGCTAT

10

005260 15139960

Figur 7. Partial DNA s-quence for the plasmid for
 insertion into yeast in which: cleotide nos. 1-173
 make the MF 1 promoter r gion and 5' noncoding
 sequence. 174-440 is th MF 1 N-terminal coding
 sequence. 441-695 is an HPTH sequence. 696-726 is an
 HPTH 3' noncoding sequ nce from pSSHPTH-10. 727-732 is
 from pUC19. 733-874 is MF 1 3' noncoding sequence and
 transcriptional termination signal.

10 10 30
 50
 AGTGCAAGAAAACCAAAAAGCAACAACAGGTTTTGGATAAGTACATATATAAGAGGGCCT
 70 90 110
 TTTGTTCCCATCAAAAATGTTACTGTTCTTACGATTCAATTTACGATTCAAGAATAGTTCA
 15 130 150 170
 AACAAGAAGATTACAAACTATCAATTTACATACACAATATAAACGACCAAAAGAATGAGAT
 190 210 230
 TTCCTTCAATTTTTACTGTCAGTTTTATTTCGCAGCATCCTCCGCATTAGCTGCTCCAGTCA
 250 270 290
 ACACTACAACAGAAGATGAAACGGCACAAATTCCGGCTGAAGCTGTCATCGGTTA[†]TCAG
 20 310 330 350
 ATTTAGAAGGGGATTTTCGATGTTGCTGTTTTGCCATTTTCCAACAGCACAAATAACGGGT
 370 390 410
 TATTGTTTATAAATACTACTATTGCCAGCATTGCTGCTAAAGAAGAAGGGGTATCTTTGG
 430 450 470
 25 ATAAAAGAGAGGCTGAAGCTWSNGTNWSNGARATHCARYTNATGCAYAAYYTNGGNAARC
 490 510 530
 AYYTNAAYWSNATGGARMGNGTNGARTGGYTNMGNAARAARYTNCARGAYGTNCAYAAYT
 550 570 590
 TYGTNGCNYTNGGNGCNCNYTNGCNCNMGNAYGCNNGNWSNCARMGNCNMGNAARA
 610 630 650
 ARGARGAYAAAYGTNYTNGTNGARWSNCAYGARAARWSNYTNGGNGARGCNGAYAARGCNG
 670 690 710
 5 AYGTTAAAYGTNYTNACNAARGCNAARWSNCARTRRAAATGAAAACAGATATTGTCAGAGT

005260" 15 FEB 96

730 750 770
TCTC GAGTCGACTTTGTTCCCACTG1 TAGCTCGTACAAAATACAATATAC
90 810 830
TTTTCAATTTCTCCGTAAACAACCTGTTTTCCCATGTAATATCCTTTTCTATTTTTCGTTT

10

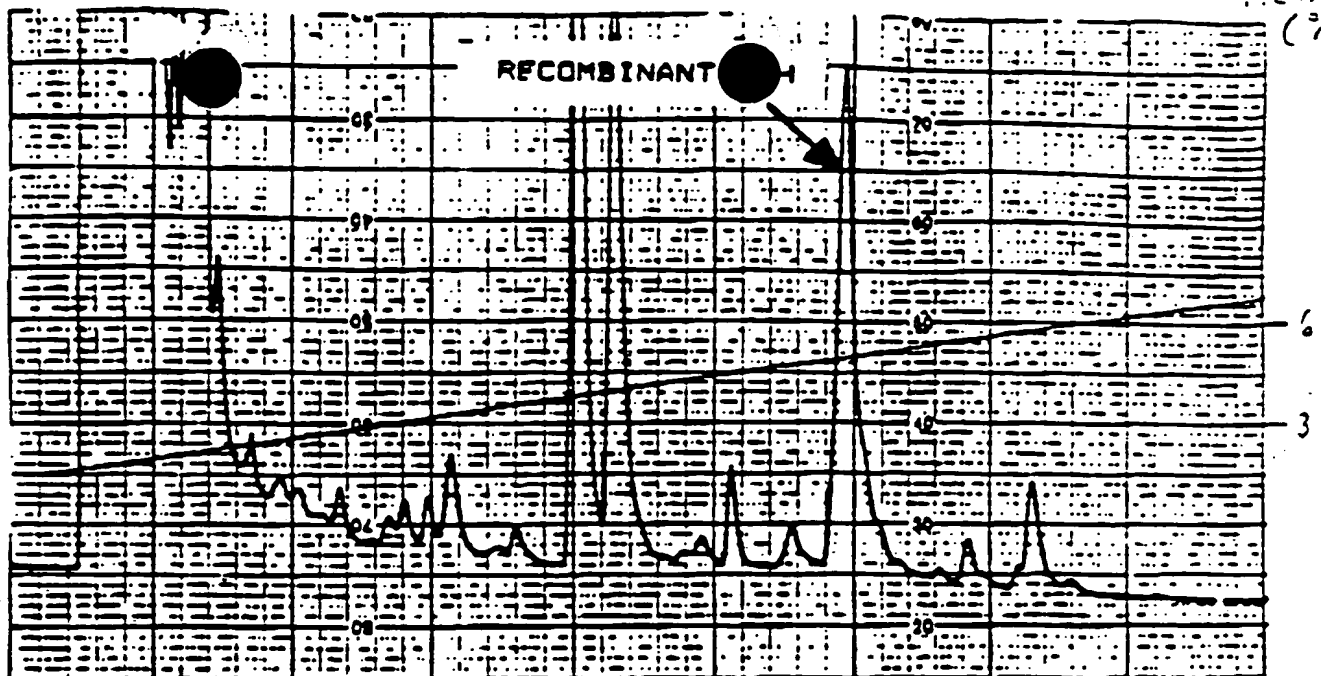
850 870
CGTTACCAACTTTACACATACTTTATATAGCTAT, wherein

15

M = A or C
R = A or G
W = A or T
S = C or G
Y = C or T
H = A or C or T
N = A or G or C or T

009260 49489560

005260-1978960



B.

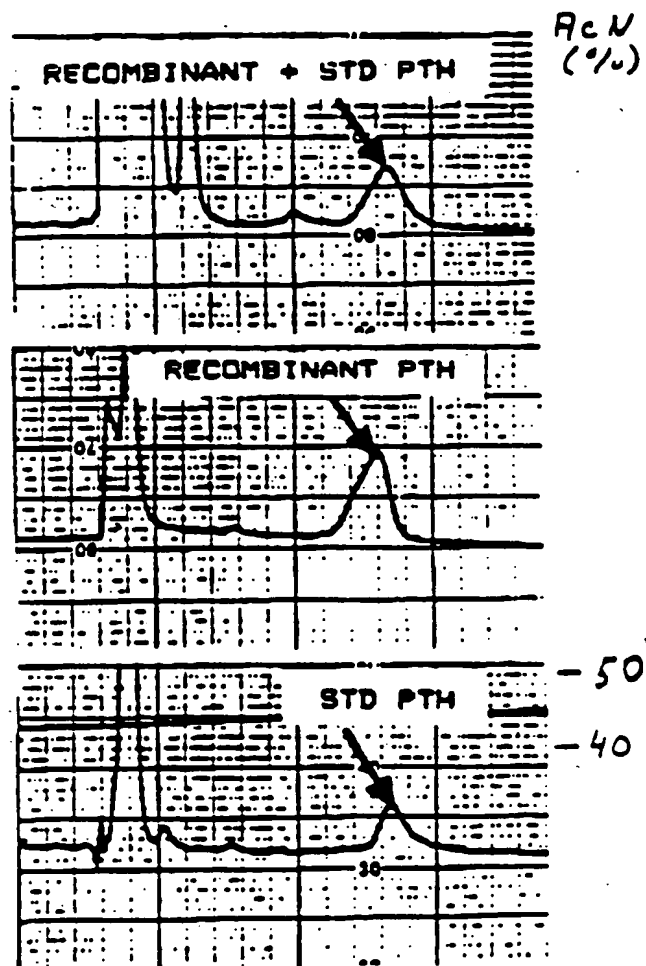


FIG. 9

FIG. 10

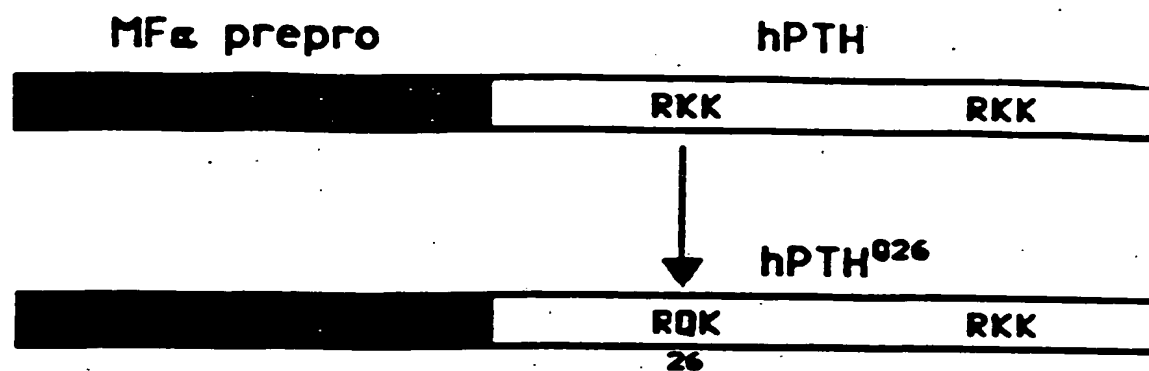


FIG. 11

005260-15139960

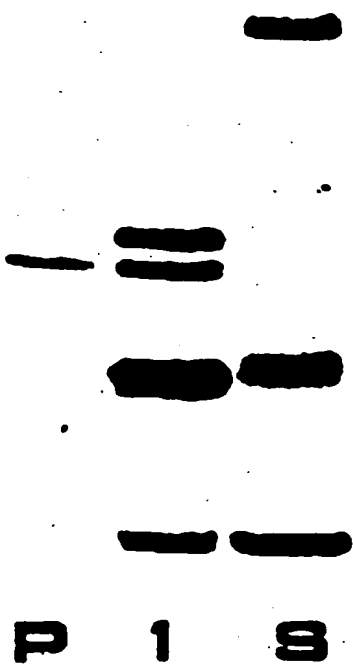
a



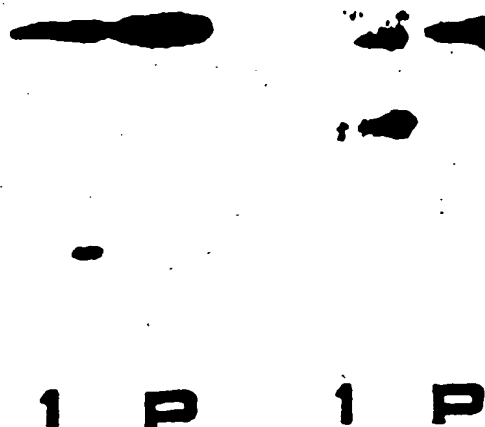
b



c



P 1 8



1 P 1 P

FIG. 8

005260-15189960

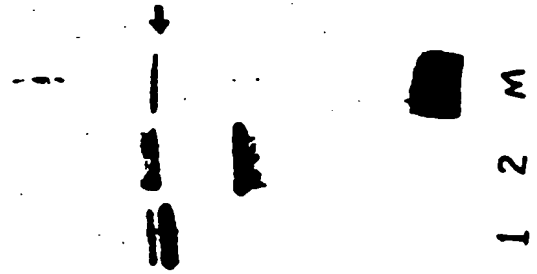
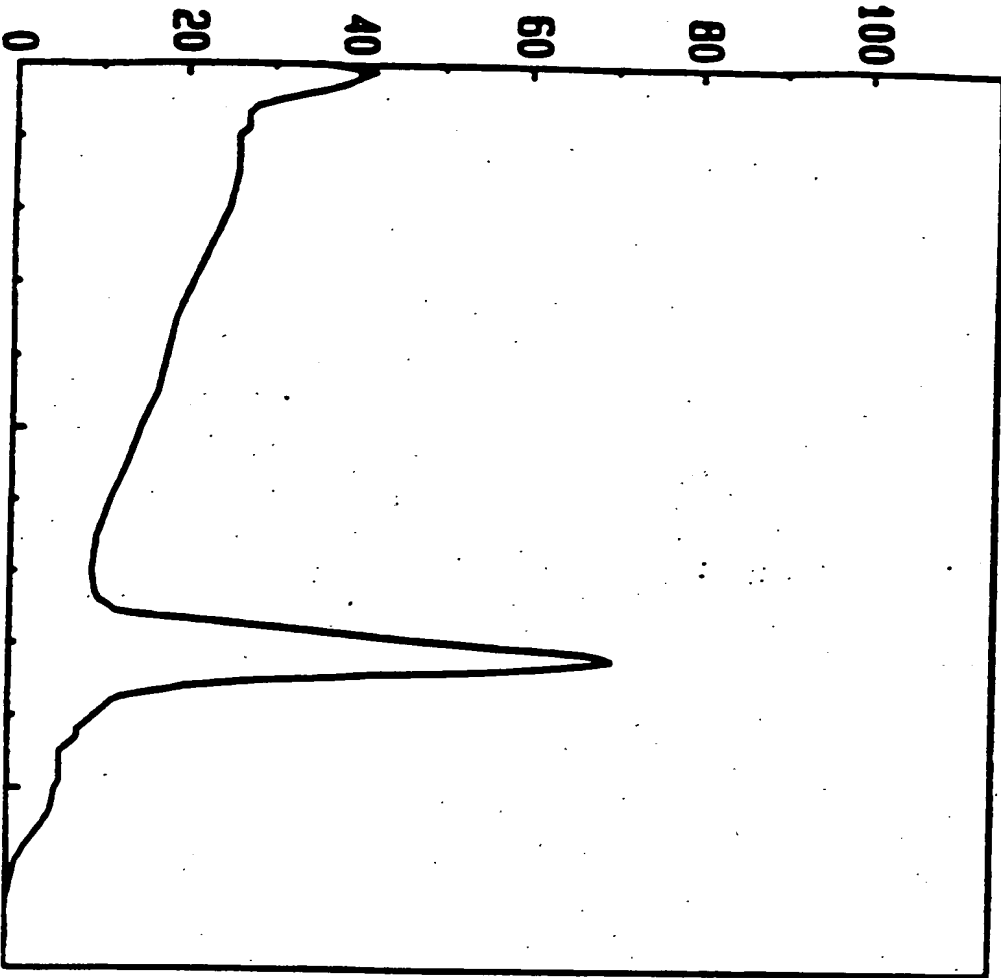


FIG. 12

A

HPLC CHROMATOGRAM OF hPTH(026)



B

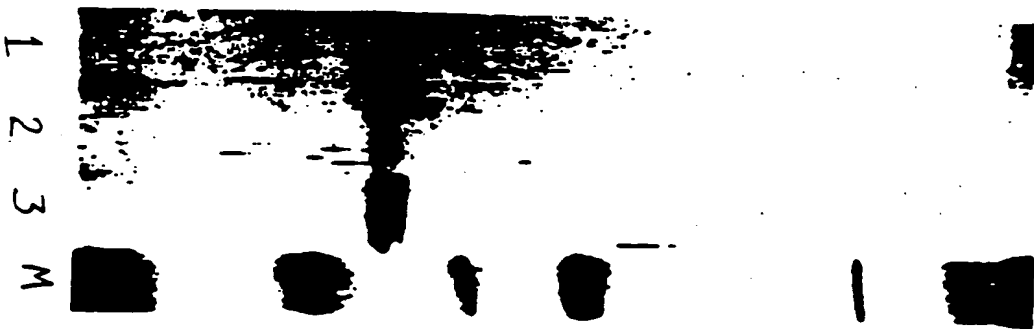


FIG. 13

Elution time

0968154.092500

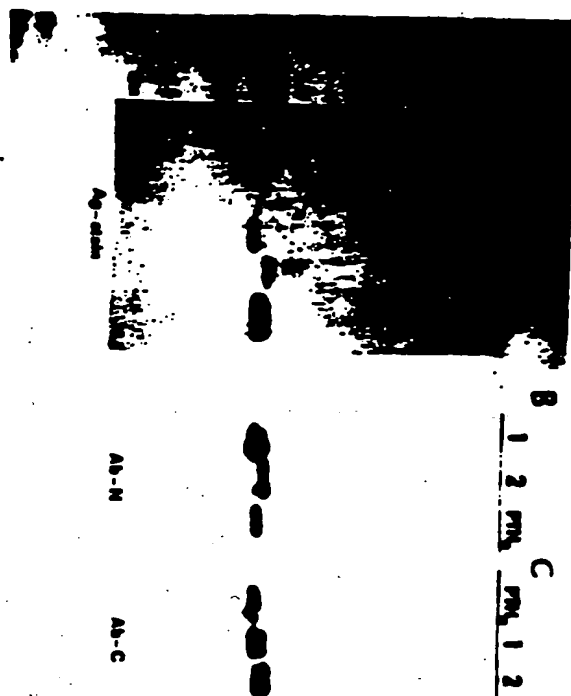
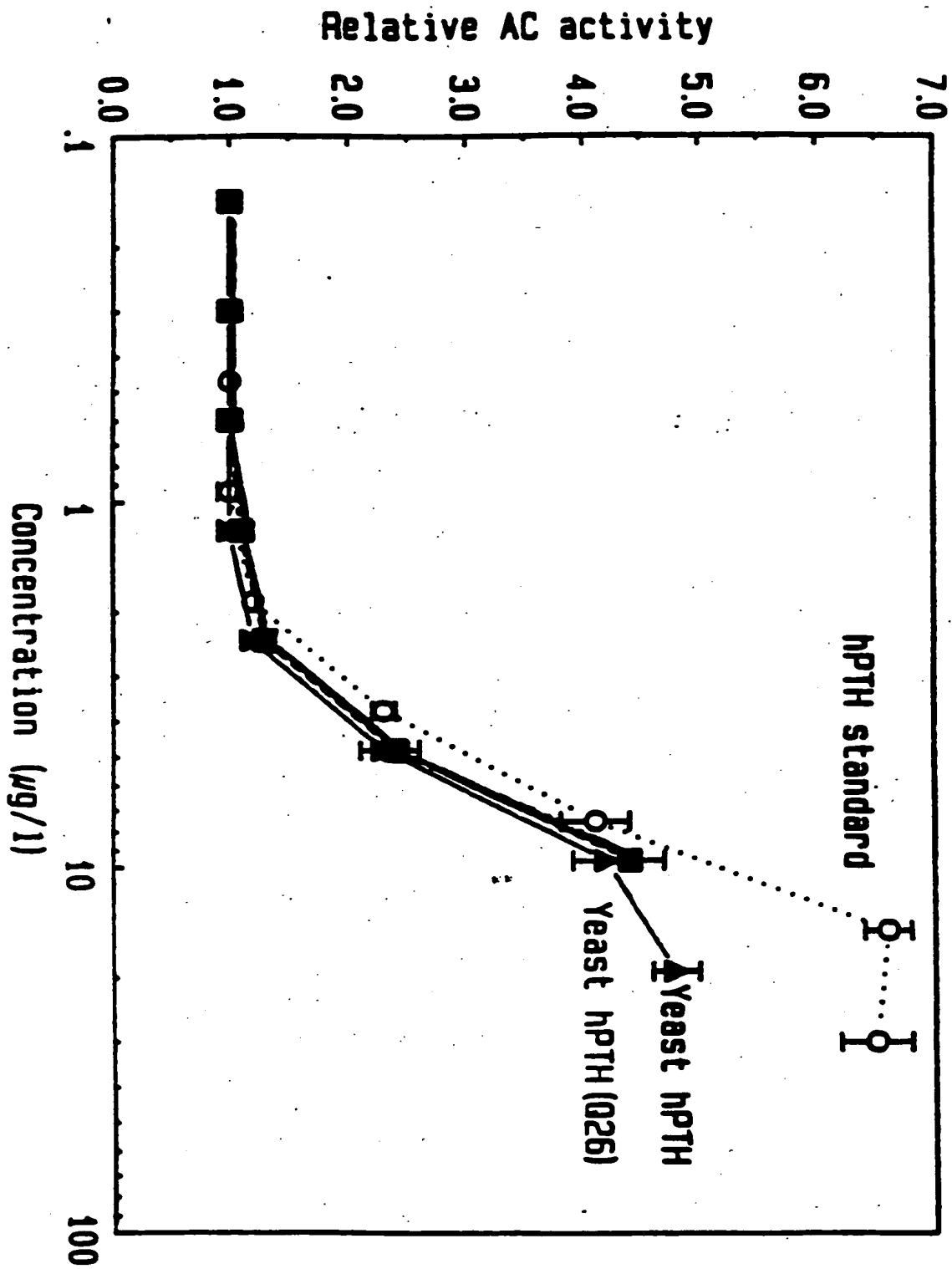


FIG. 14

FIG. 15



0968454.092500